

Soil Moisture Tension and Microbiological Activity¹

H. D. BHAUMIK AND FRANCIS E. CLARK²

MOISTURE is recognized as essential for the biological transformation of complex organic material in soil. Previous investigators (1, 3, 5, 8)³ have considered the influence of differing amounts of moisture on the activity of soil microorganisms as measured by microbial numbers, by evolution of carbon dioxide, by the formation of ammonia or nitrates, or by the fixation of elementary nitrogen. For such work, moisture content has been expressed in terms of percentage of maximum water-holding capacity or in terms of the thickness of the moisture film. To the experimental difficulty of separating the effects of moisture from other inter-acting factors, there has been added in some instances failure to insure a constant moisture content throughout an experimental period or to effect an even distribution of moisture throughout the sample.

During recent years, the energy concept of soil moisture has been more widely developed. Under this concept, soil moisture is expressed in terms of the physical forces by which it is held in soil and not in terms of actual percentage content. The present study has been undertaken to consider the effect of soil moisture tension on microbiological activity, as measured by carbon dioxide evolution, under controlled conditions of incubation.

EXPERIMENTAL

Five soil lots, selected because of differences in texture, were collected from cultivated fields near Ames, Iowa. Prior to use, each lot was air dried and screened to pass a 2-mm sieve.

Moisture contents for tensions of "0" and "1" cm of water were determined by the use of standard moisture equivalent boxes. In each instance, 30 grams of air-dried screened soil were placed on filter paper within the box, levelled by gentle tapping, and the box placed overnight in a tray containing distilled water slightly above the surface of the soil. The next day the boxes were allowed to drain by gravity for 30 minutes, with the lids in place. For "0" cm of tension, excess moisture on the bottom was wiped but once with a paper towel. For "1" cm of tension, wiping was performed with blotting paper several times, or until no more water drops appeared. Moisture contents established by these two differing procedures are shown in Table I.

Moisture contents for 10 and 50 cm of water tension were determined manometrically.

Moisture content at the "moisture equivalent", or 502 cm of water tension, was determined by the centrifuge method (6) and at the permanent wilting percentage (15,340 cm of tension) by growing sunflowers in suitable containers (4). Moisture content for 3,160 cm of water tension was determined by interpolation from curves constructed from data obtained in the several determinations just cited.

Total carbon for the several soils was determined according to the recommendations of Salter (7) and Winter and Smith (9), with minor modifications.

¹Soils Subsection, Iowa Agricultural Experiment Station, Ames, Iowa, and Division of Soil Management and Irrigation, Bureau of Plant Industry, Soils, and Agricultural Engineering, Agricultural Research Administration, U. S. Dept. of Agriculture, co-operating. Journal Paper No. J-1477, Project 956, of the Iowa Station.

²Graduate Student, Iowa State College, and Bacteriologist, Bureau of Plant Industry, Soils, and Agricultural Engineering, respectively.

³Figures in parenthesis refer to "Literature Cited", p. 238.

TABLE I.—Moisture contents determined for five Iowa soils at selected tension values.

Soil type	Percentage of moisture at cm of water tension of						
	0	1	10	50	502	3,160	15,340
Thurman sand	43.69	34.31	26.86	14.12	8.98	5.6*	3.04
Clarion loam	59.89	51.60	36.69	25.63	17.57	11.7	7.04
Clarion fine sandy loam	61.34	53.47	38.85	30.59	17.66	11.8	7.16
Webster silt loam	76.40	60.07	47.55	34.11	24.03	16.5*	10.52
Wabash silty clay	93.11	76.40	58.55	42.25	28.79	21.1	15.09

*Values for this column by interpolation.

For establishing incubation experiments at the lower tensions, soil lots could be brought to the desired moisture contents simply by adding water from a burette. For the higher moisture tensions of 502 cm and 3,160 cm, corresponding to pF of 2.7 and 3.5 on the assumption that the soil moisture energy is entirely accounted for by the curvature of the moisture films, a known weight of air-dry soil was spread evenly on a large enameled tray and the amount of water required was applied by spraying, with the use of compressed air, in five or six instalments, the sample being mixed thoroughly with a spatula after each instalment to insure uniform moisture distribution throughout the sample.

Incubation flasks received soil equivalent to 100 grams, oven-dry basis. One per cent of finely ground corn stover was thoroughly mixed in all soil lots prior to watering. Incubations were made at 30° C, $\pm 1^\circ$. 500-ml pyrex Erlenmeyer flasks generally were used as incubation containers. In a few instances to be noted below, flint glass bottles approximately 5 \times 5 \times 15 cm in size were used. Incubation containers were fitted with 2-hole rubber stoppers and connected with an air stream maintained at constant pressure by means of a water column. The air stream was passed through concentrated potassium hydroxide solution, concentrated sulfuric acid, soda lime, ascarite, and calcium chloride towers, and finally, through a series of long, horizontal tubes half-filled with distilled water to saturate the air stream to 100% relative humidity before it entered the incubation containers.

The outlet tubes carrying the gases from the incubated soils were led through holes in the side of the incubator and connected to tubes containing standard sodium hydroxide. Contents of the collecting tubes were analyzed after 1, 2, 3, 4, 6, 9, 12, and 15 days for carbon dioxide, according to standard methods.

Microbiological analyses by cultural procedures (2) were limited to determinations of the numbers of fungi, actinomycetes, and bacteria in selected series of containers.

OBSERVATIONS

The moisture/tension relationships of the five soils employed are shown in Table I. It may be seen that the capacity for retention of water against a given tension progressively increases in the following order: Thurman sand < Clarion loam < Clarion fine sandy loam < Webster silt loam < Wabash silty clay.

Total carbon was determined present in the five soils as follows: Thurman sand, 0.75%; Clarion loam, 1.47%; Clarion fine sandy loam, 1.75%; Webster silt loam, 2.93%; Wabash silty clay, 2.92%.

Total amounts of carbon dioxide evolved from the several soils during 15 days of incubation at differing moisture contents are shown in Table 2.

It is apparent that for the several soils there is no regularity between total carbon dioxide evolved during the 15-day period and moisture content. In fact, no two of the five soils showed maximum carbon dioxide production at the same tension value. The lighter textured soils showed maxima at the higher tensions; the heavy-textured, at the lower tensions.

In Webster silt loam, cumulative total carbon dioxide evolved increased up to 1 cm of tension, and then diminished with a further increment in moisture. In Wabash silty clay, the maximum occurred at 10 cm of tension. As the deleterious effect of increased moisture beyond a certain maximum is believed generally to be due to its limitation of aeration, a supplementary experiment was performed with Webster silt loam incubated in dilution bottles rather than in Erlenmeyer flasks. In this manner a much smaller surface/volume ratio was established, but the moisture tension values employed remained unchanged. The cumulative totals of carbon dioxide evolved in 15 days from the two types of containers are compared in Table 3. It may be seen that up to 10 cm of water tension, there is no reduction in activity due to reduction in surface/volume ratio, whereas at 1 and at 0 cm of tension, there is less carbon dioxide evolved from the containers with decreased surface and increased depth of soil column.

Although no definite trend in the behavior of the five soils was apparent from total amounts of carbon dioxide evolved, rates of evolution per 24-hour period reveal in all cases that peak rates are attained at or near a moisture tension equivalent to 50 cm of water. Table 4 presents the peak rates of carbon dioxide evolution encountered at each of six moisture tensions during the course of 15-day incubations of the five different soils. As this table gives only the peak evolution per 24 hour period, Fig. 1 is presented to show the rate of evolution throughout the course of 15 days for one soil type, Webster silt loam. Curves constructed for the remaining four soils were essentially similar to those depicted in Fig. 1.

TABLE 2.—Total carbon dioxide evolution during 15 days from soils maintained at differing moisture tensions.

Soil type	Mgm carbon dioxide during 15 days incubation with cm of water tension of					
	0	1	10	50	502	3,160
Thurman sand . . .	343.25*	327.46	446.99	507.04	508.02	563.15
Clarion loam	322.99	327.81	316.43	312.91	360.03	299.07
Clarion fine sandy loam	380.16	393.99	418.89	503.96	446.53	416.86
Webster silt loam . . .	431.32	513.98	426.28	408.75	381.49	370.55
Wabash silty clay . . .	421.68	412.32	449.56	397.20	332.35	322.34

*Each value given is the mean of two replicates.

TABLE 3.—Comparative carbon dioxide evolution from Webster silt loam incubated at differing moisture tensions in shallow and deep containers.

Cm of water tension	Mgm of carbon dioxide during 15 days incubation	
	Soil in shallow containers	Soil in deep containers
0	431.32*	188.67
1	513.98	409.36
10	426.28	501.49
50	408.75	450.91
502	381.49	397.08
3,160	370.55	344.32

*Each value given is the mean of two replicates.

For all soils, curves of CO_2 evolution at the high moisture tensions showed rapid increase in rates during the initial days of incubation, followed by a rapid decrease. These curves show a logarithmic ascent, followed by a similar descent, such as is characteristic of the growth curve of microorganisms.

TABLE 4.—Peak rates of carbon dioxide evolution encountered at differing moisture tensions.

Cm of water tension	Peak rate, in mgm CO_2 per 24 hours	Time of occurrence during the 15 days of incubation, day
Thurman Sand		
0	24.22	2nd
1	26.92	2nd
10	75.18	2nd
50	85.83	2nd
502	86.47	3rd
3,160	85.66	4th
Clarion Loam		
0	22.75	4th
1	26.38	2nd
10	54.33	1st
50	66.88	1st
502	71.25	1st
3,160	46.56	2nd
Clarion Fine Sandy Loam		
0	31.84	4th to 6th
1	33.95	4th to 6th
10	51.64	1st
50	81.97	1st
502	68.20	2nd
3,160	60.79	2nd
Webster Silt Loam		
0	32.14	4th to 6th
1	36.35	2nd
10	73.82	1st
50	78.45	1st
502	59.60	3rd
3,160	52.64	3rd
Wabash Silty Clay		
0	32.61	6th to 9th
1	29.22	2nd
10	69.11	1st
50	74.84	1st
502	49.57	2nd
3,160	45.02	2nd

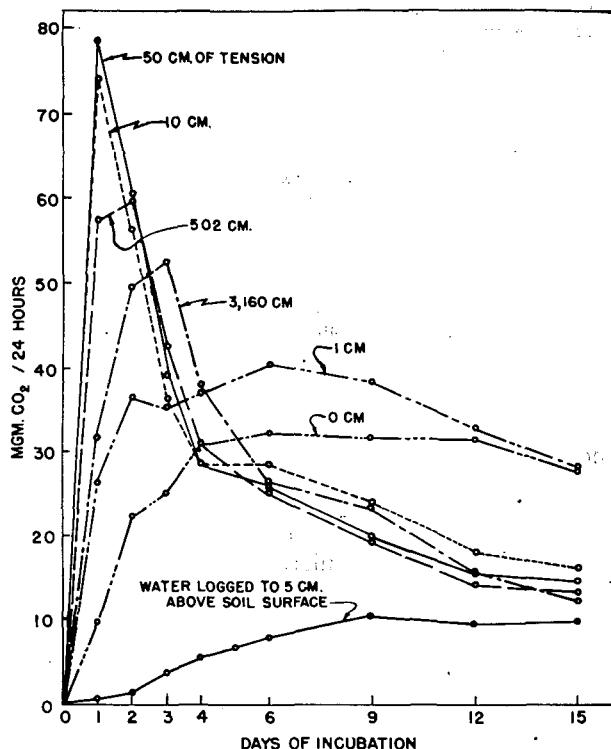


FIG. 1.—Rate of carbon dioxide production in Webster silt loam incubated 15 days at selected moisture contents. Numbers near peaks of the individual curves represent centimeters of water tension during incubation.

At or near saturation, curves present a different appearance. The initial sharp peak is absent; there appears rather to be two peak rates, one during the initial stage of incubation and another at approximately 6 to 9 days.

From Table 4, it may be seen that there is a tendency for all soils to show their maximum peak rate at 50 cm of tension. In the case of Clarion fine sandy loam, Webster silt loam, and Wabash silty clay, the rate at 50 cm of tension was distinctly maximal. In the case of Clarion loam, the peak rate was at 502 cm, but the excess over the rate at 50 cm was very slight. For Thurman sand, there was no material difference between the rates at 50, 502, and 3,160 cm of tension.

Microbiological analyses revealed differences in the abundance of microbial groups not only between soils differing in texture but also between differing tensions of moisture within the same soil. Numbers of fungi, actinomycetes, and bacteria enumerated for five soil types after 5 and 15 days of incubation at moisture tensions equivalent to 1 cm and to 3,160 cm of water are shown in Table 5.

At the higher moisture tension, the maximum number of fungi was observed in the coarse textured and the maximum number of bacteria in the fine-textured soil.

Sufficient replicates were available on the Webster silt loam incubated in bottles, as described above, to

TABLE 5.—Microbial numbers determined after 5 and 15 days of incubation at moisture tensions equivalent to 0 and 3,160 centimeters of water.

Soil type	Fungi, thousands per gram		Actinomycetes, millions per gram		Bacteria, millions per gram	
	5 days	15 days	5 days	15 days	5 days	15 days
At 0 cm Tension						
Thurman sand.....	63 63	130 115	18 14	43 30	58 61	54 33
Clarion loam.....	37 17	31 27	10 15	29 39	122 77	32 25
Clarion fine sandy loam	10 4	21 22	11 12	38 33	128 90	45 33
Webster silt loam.....	4 10	20 25	14 17	37 41	64 88	34 43
Wabash silty clay.....	13 3	56 43	15 14	54 40	51 84	57 50
At 3,160 cm Tension						
Thurman sand.....	6,000 7,600	11,700 10,400	18 34	30 20	81 88	38 26
Clarion loam.....	243 240	420 370	63 50	72 57	145 122	113 85
Clarion fine sandy loam	127 147	320 380	33 43	133 70	280 296	220 230
Webster silt loam.....	233 197	620 610	47 39	62 77	143 138	148 167
Wabash silty clay.....	150 167	620 730	38 34	93 70	316 337	360 297

permit microbiological analyses. The data accumulated are given in Table 6.

TABLE 6.—Microbial numbers determined for Webster silt loam following 5 and 15 days of incubation at selected moisture tensions.

Moisture tension equivalent in cm of water	Fungi, thousands per gram		Actinomycetes, millions per gram		Bacteria, millions per gram	
	5 days	15 days	5 days	15 days	5 days	15 days
0	25 26	25 39	7.3 7.7	6.5 3.5	16 11	28 21
1	26 21	40 34	7.0 5.0	4.0 3.7	12 9	26 18
10	35 31	74 73	13.3 7.0	8.3 7.0	66 63	60 30
50	58 62	287 250	10.0 8.0	8.0 4.0	97 82	70 66
502	113 104	463 517	41.5 38.8	28.0 27.0	85 131	105 110
3,160	107 110	593 517	76.1 55.0	67.0 50.0	105 140	97 110

In the final experiment here recorded, six replicates each of Thurman sand and of Wabash silty clay were established in Erlenmeyer flasks. In each series, four containers were established at 50 cm and two at 0 cm of tension. The latter, and two of the former, were incubated undisturbed for 15 days. The remaining two containers, established at 50 cm of tension, received sufficient water after 48 hours of incubation to bring them to 0 cm of tension, at which moisture content they were incubated for the next 13 days. Carbon dioxide evolved from the individual flasks was determined at daily to 3-day intervals. The data are shown in Table 7.

Daily rates of carbon dioxide evolution from the two soils are shown in Table 8. Fig. 2, constructed from data obtained on Wabash silty clay, shows the abrupt shift from the type of curve typical of 50 cm of tension to the type of curve typical of 0 cm of tension (Fig. 1).

DISCUSSION

Considering the cumulative totals of carbon dioxide evolved, there appears a difference between sandy soils, on the one hand, and silt loam and silty clay, on the other. The observation that at a given moisture tension maximum number of fungi were found in

TABLE 7.—*Cumulative total carbon dioxide evolved in mgm from soils at constant and at abruptly changed moisture contents.*

Soil type	50 cm tension for 15 days	50 cm tension for 2 days, then 0 cm for 13 days	0 cm tension for 15 days
Thurman sand....	507.04	381.80	343.25
Wabash silty clay	415.44	400.07	421.68

TABLE 8.—*Rates of carbon dioxide evolution in mgm per 24 hours from soils at constant and with abruptly changed moisture contents.*

Day of incubation	At 50 cm tension for 15 days	At 50 cm for 2 days, then at 0 cm tension for next 13 days	At 0 cm tension for 15 days
Thurman Sand			
1	56.18	56.18	6.91
2	85.83	85.83	24.22
3	70.54	32.61	22.10
4	55.50	23.16	21.09
6	42.41	17.55	23.93
9	23.29	14.45	26.80
12	14.62	18.06	26.78
15	13.49	17.13	18.64
Wabash Silty Clay			
1	54.59	54.59	6.24
2	64.87	64.87	16.07
3	52.34	11.94	25.40
4	38.40	13.59	29.30
6	27.89	21.09	32.32
9	17.17	25.56	32.61
12	17.27	22.60	31.12
15	15.04	22.80	27.03

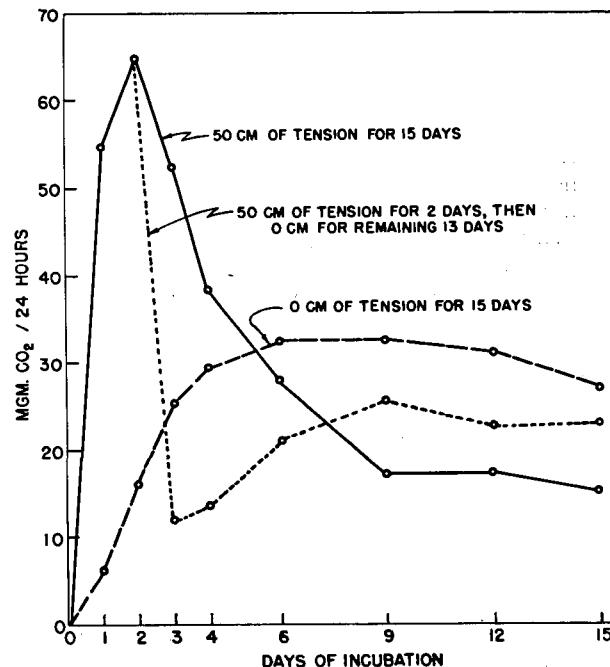


FIG. 2.—Rates of carbon dioxide evolution from Wabash silty clay during 15 days of incubation at constant and at abruptly changed moisture content.

Thurman sand and maximum number of bacteria in Wabash silty clay suggests that soil texture not only determines the quantity of water that can be retained against a particular tension but also influences the nature of the soil microflora, thereby affecting the course of microbiological decomposition as revealed by carbon dioxide evolution.

When peak rates of evolution of carbon dioxide rather than total production are considered, there is a general tendency for all the soils employed to show maximum rates of evolution at their respective moisture contents equivalent to 50 cm of water tension. The peak rate may be interpreted as indicative of the conditions most favorable for carbon dioxide production by the soil microflora. At a tension of 50 cm of water, by convention "the aeration porosity limit", it is probable that the general microbial population is provided with the most favorable aeration condition that can exist simultaneously with maximum thickness of capillary moisture film.

Differences noted in carbon dioxide evolution from wet soils following alteration of the surface/volume ratio by use of differently shaped containers emphasize the importance of aeration on the activity of aerobic microorganisms in wet soil. In the case of comparatively dry soil, the surface/volume ratio appeared much less important.

SUMMARY

Five Iowa soils differing in texture, but all receiving 1% finely ground corn stover, were incubated under standard conditions at moisture tensions of 3,160, 502, 50, 10, 1, and 0 cm of water. Carbon

dioxide evolved was collected continuously and was determined after 1, 2, 3, 4, 6, 9, 12, and 15 days.

Moisture contents of the various soils at the tensions named, total amounts of carbon dioxide evolved from the five soils at each tension, and the peak rates of evolution (per 24 hour period) are recorded.

The moisture tension at which the maximum amount of carbon dioxide was evolved during 15 days differed for the several soils. For Thurman sand, this tension was found to be in the vicinity of 3,160 cm of water. For Clarion loam, it was 502 cm; for Clarion fine sandy loam, 50 cm; for Wabash silty clay, 10 cm; and for Webster silt loam, 1 cm.

For all soils, the peak rate of carbon dioxide production was observed at or very near to the moisture tension at the aeration porosity limit, taken by convention at 50 cm of water. Sand was exceptional in that there was little difference between the rates at 50, 502, and 3,160 cm of tension.

Microbiological analyses revealed differences in the abundance of microbial groups both at differing tensions and for soils differing in texture. It is believed that such differences in microbial populations are at least partly responsible for differences in the amounts of carbon dioxide evolved from the several soils.

LITERATURE CITED

1. BOLLEN, W. B. Soil respiration studies on the decomposition of native organic matter. *Iowa State Col. Jour. Sci.*, 15: 353-374. 1941.
2. CLARK, FRANCIS E. Notes on types of bacteria associated with plant roots. *Kans. Acad. Sci. Trans.*, 43: 75-84. 1940.
3. GREAVES, J. E., and CARTER, E. G. Influence of moisture on the bacterial activities of the soil. *Soil Sci.*, 10: 361-387. 1920; 13: 251-270. 1922.
4. HENDRICKSON, A. H., and VEIHMEYER, F. J. Permanent wilting percentage of soils obtained from field and laboratory trials. *Plant Physiol.*, 20: 517-539. 1945.
5. RAHN, O. Bacterial activity in soil as a function of grain size and moisture content. *Mich. Agr. Exp. Sta. Tech. Bul.* 16. 1912.
6. RICHARDS, L. A., and WEAVER, L. R. Fifteen atmosphere percentage as related to permanent wilting percentage. *Soil Sci.*, 56: 331-340. 1943.
7. SALTER, R. M. A rapid method for the accurate determination of total carbon in soils. *Jour. Ind. and Eng. Chem.*, 8: 637-639. 1916.
8. WAKSMAN, S. A. *Principles of Soil Microbiology*. Baltimore: Williams & Wilkins. 1927.
9. WINTER, E., JR., and SMITH, R. S. Determination of total carbon in soils. *Ind. and Eng. Chem., Anal. Ed.*, 1: 202-203. 1929.